# Effect of the Insecticide Zectran (Mexacarbate) on Several Algae<sup>1</sup>

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An intensive search has been conducted for insecticides which lack the devastating properties of DDT in that they are neither persistent nor toxic to fish and wildlife. Zectran<sup>(R)</sup> Mexacarbate (4-dimethylamino - 3,4-xylyl methylcarbamate) degrades rapidly in sunlight (ABDEL-WAHAB & CASIDA 1967) and its effects on mule deer and avians (TUCKER & CRABTREE 1969), soil microbes (BOWEN <u>et al</u>. Unpub.) and both terrestrial (ROBERTS, <u>et</u> <u>al</u>. 1969) and aquatic insects (GIBSON & CHAPMAN 1972) have been studied. A knowledge of Zectran's effect on aquatic primary producer communities was lacking at the time it was registered for use on certain forest insects.

In a previous study we determined (SNYDER & SHERIDAN 1974) that Zectran exhibited a toxicity threshold between  $10^3$  to  $10^4$  ppb on four laboratory strains of algae. In this study the effects of Zectran are determined for several additional algal genera collected in the field and either returned to the laboratory for testing or tested under field conditions.

# MATERIALS AND METHODS

# Experimental

Zectran, as formulated by Dow Chemical, is dissolved in a petroleum carrier and designated FS-15. FS-15 was homogenized in stream water from the collection site and diluted to concentrations between 10 and  $10^4$  ppb. This method closely resembles the natural entry of Zectran into an aquatic system. The questionable practice of employing an organic solvent as a carrier was avoided. Experimental cells were returned to the lab from the field and incubated in several concentrations of Zectran for 1, 24 or 48 hours at  $17^{\circ}$ C at 1000 ft-c. (cool white, fluorescent). Photosynthesis and respiration rates were determined using the diethanolamine technique as described previously (SNYDER & SHERIDAN 1974).

Rates of NaHC<sup>14</sup>O<sub>3</sub> uptake were determined under field conditions by placing a suspension of the experimental cells in stream water into a 6 ml screw-capped vial along with 3 ml of a Zectran solution and 0.1 ml of 0.2 <u>M</u> carbonate-bicarbonate buffer mixture #11 (UMBREIT <u>et al</u>. 1964). The vial was then placed on its side in the water for 10 minutes to allow for thermal equilibration,

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after which 0.2 ml of NaHC<sup>14</sup>O<sub>3</sub> (1  $\mu$ c/ml) was injected and the vial returned to the water. The incorporation of tracer occurred for an incubation time of 30 minutes, and then photosynthesis was arrested by injecting 1 ml of 6 M acetic acid. This field method has the advantage that the samples are exposed to tracer at field temperatures and natural light intensities (BROCK & BROCK 1967). Samples were then returned to the laboratory, placed in 20 ml scintillation vials, evaporated to dryness at 50°C and then 15 ml of scintillation mixture added which contained 0.3% PPO and 0.01% PoPOP in toluene. The mean counts-per-minute (CPM) for each sample was determined in a liquid scintillation counter.

Chlorophyll was extracted and the concentration determined in methanol using the formulae of HOLDEN (1965).

#### Habitat

Jerry Johnson Hot Spring -- thermal springs located in Northern Idaho 30 miles south of the Montana border on Highway 12 - <u>Chroococcus</u> sp.

Pattee Creek -- small creek approximately 1 m in width draining Pattee Canyon south of Missoula - <u>Spirogyra</u> sp., <u>Ulothrix</u> sp.

Rattlesnake Creek -- 7 m wide draining Rattlesnake Canyon north of Missoula - <u>Schizogonium</u> sp., <u>Mougeotia</u> sp., <u>Vaucheria</u> sp., <u>Zygnema</u> sp.

Warm Springs Creek -- 4 m wide receives effluent from Jerry Johnson Hot Spring and is confluent with the Lochsa River in Northern Idaho - <u>Oedogonium</u> sp.

The samples were selected from large, nearly homogenous populations having a single dominant genus with other genera relatively infrequent in occurrence within the sample.

## RESULTS AND DISCUSSION

The data showing the effect of several concentrations of Zectran after 1, 24 or 48 hours of exposure to the insecticide are presented in Fig. 1. Oxygen production by <u>Mougeotia</u>, <u>Spirogyra</u> and <u>Schizogonium</u> was not inhibited by 1 hour exposure to Zectran at several concentrations between 10 and  $10^4$  parts per billion (ppb), whereas after 24 hours exposure, all three species exhibited a threshold for reduced 0<sub>2</sub> production between  $10^3$  and  $10^4$  ppb. A sample composed of <u>Spirogyra</u>, <u>Mougeotia</u> and <u>Zygnema</u>, designated mixture, showed a depressed 0<sub>2</sub> production by 30 percent of the control after 1 hour of exposure to  $10^4$  ppb Zectran and 50% after 48 hours of exposure to the same concentration.

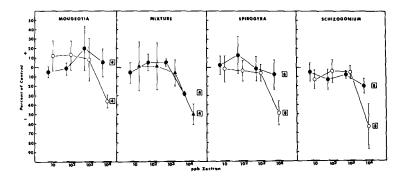


Fig. 1. The mean rate of net oxygen production of six genera of algae in several concentrations of Zectran. The cells were exposed to the various concentrations for 1 (o - o), 24 ( $\bigcirc$  -  $\bigcirc$ ) or 48 ( $\land$  -  $\land$ ) hours. The number of trials is shown  $\Box$ . Vertical bars show confidence limits at the 95% level of significance.

The rate of  $C^{14}O_2$  (Fig. 2) incorporation by both <u>Oedogonium</u> and <u>Chroococcus</u> as measured under field conditions was reduced by exposure to Zectran at a concentration of  $10^4$  ppb for 1 hour.

The mean photosynthesis values shown in Figures 1 and 2 for Zectran concentrations between 10 and  $10^3$  ppb showed a tendency toward stimulation or inhibition as compared to the control. However, these were not different from the control at 95% level of significance (SOKAL & ROHLF 1969).

In a previous paper (SNYDER & SHERIDAN 1974), we determined that Zectran exhibited a threshold for toxicity on growth between  $10^3$  and  $10^4$  ppb in laboratory cultures of <u>Scenedesmus quadricauda</u>, <u>Navicula pelliculosa</u>, <u>Synechococcus lividus strain Y52-s and Oscillatoria terebriformis strain OH-51</u>. However, the insecticide Zectran decreased both photosynthesis and respiration in <u>O. terebriformis and S. lividus at concentrations between 0.5 and 12.5</u> parts per million (ppm) (mg/1) whereas <u>N. pelliculosa and S. quadricauda</u> were unaffected by these same concentrations in these short-term manometric measurements.

It was important to continue these studies to determine the threshold for toxicity for species sampled from natural habitats. The samples were either returned directly to the laboratory where photosysnthesis was determined by standard manometric techniques or the rate of  $C^{14}O_2$  uptake was determined in the field.

The results showed that  $0_2$  production by field collections of <u>Mougeotia</u>, <u>Spirogyra</u> and <u>Schizogonium</u> was reduced at 24 hours of exposure to Zectran at a concentration of  $10^4$  ppb. Oxygen production by the sample designated mixture and  $C^{14}O_2$  uptake by <u>Chroococcus</u> and <u>Oedogonium</u> were reduced after 1 hour exposure to Zectran at a concentration of  $10^4$  ppb.

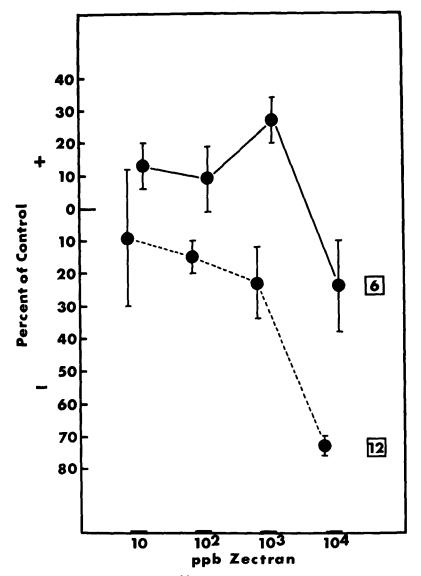


Fig. 2. The mean rate of  $C^{14}O_2$  photoincorporation by two algal genera after 1 hour of exposure to several concentrations of Zectran. The number of trials is shown  $\Box$ . Vertical bars show confidence limits at the 95% level of significance. Chroococcus sp. \_\_\_\_; Oedogonium sp. -----.

The final concentration of Zectran in various field situations is influenced by the concentration of insecticide introduced and the volume of the aquatic system which in certain circumstances is determined by the extent of mixing within the epilimnion and/or the degree of mixing within the basin. In order to extrapolate these results to the field situation, we calculated the concentration of Zectran resulting from the direct spraying of a lake 4046 m<sup>2</sup> by 1 m deep with Zectran at the registered concentration of 68 g per acre. This would result in a Zectran concentration of 16.6 ppb which was approximately 100fold less than that required to effect a decrease in growth in the algal species tested.

#### SUMMARY

Field samples of freshwater algae were examined to determine the effect of the insecticide Zectran on photosynthesis rate. Concentrations of Zectran between 10 and  $10^3$  parts per billion (ppb) affected neither  $0_2$  production nor NaH<sup>14</sup>CO<sub>3</sub> uptake in any of the seven genera tested. However, Zectran at a concentration of  $10^4$  ppb after 1 hour effected a reduction in photosynthesis of <u>Chroococcus, Oedogonium</u> and in a mixed sample composed of <u>Zygnema</u>, <u>Mougeotia and Spirogyra</u>. <u>Mougeotia</u>, <u>Spirogyra and Schizogonium</u> were not affected by exposure to  $10^4$  ppb Zectran after 1 hour, but  $0_2$  production was significantly reduced after 24 hours of exposure to this same concentration.

#### ACKNOWLEDGMENT

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